PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 0100WO00ORD	**FOR FURTHER ACTION**	See Form PCT/IPEA/416
International application No. PCT/EP2005/050739	International filing date *(day/month/year)* 21.02.2005	Priority date *(day/month/year)* 23.02.2004
International Patent Classification (IPC) or national classification and IPC INV. C12N7/02 C12N7/00		
Applicant CRUCELL HOLLAND B.V. et al.		

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of 8 sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
   a. ☐ *sent to the applicant and to the International Bureau)* a total of sheets, as follows:
      ☐ sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
      ☐ sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.
   b. ☐ *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in electronic form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).

4. This report contains indications relating to the following items:
   - ☒ Box No. I Basis of the report
   - ☐ Box No. II Priority
   - ☐ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - ☒ Box No. IV Lack of unity of invention
   - ☒ Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - ☐ Box No. VI Certain documents cited
   - ☐ Box No. VII Certain defects in the international application
   - ☐ Box No. VIII Certain observations on the international application

Date of submission of the demand	Date of completion of this report
23.01.2006	18.07.2006
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized officer Herrmann, K Telephone No. +49 89 2399-2670

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/EP2005/050739

Box No. I Basis of the report

1. With regard to the **language**, this report is based on

   ☒ the international application in the language in which it was filed

   ☐ a translation of the international application into , which is the language of a translation furnished for the purposes of:

   - ☐ international search (under Rules 12.3(a) and 23.1(b))
   - ☐ publication of the international application (under Rule 12.4(a))
   - ☐ international preliminary examination (under Rules 55.2(a) and/or 55.3(a))

2. With regard to the **elements*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

   Description, Pages

   1-56 as originally filed

   Claims, Numbers

   1-32 as originally filed

   Drawings, Sheets

   1/12-12/12 as originally filed

   ☒ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing

3. ☐ The amendments have resulted in the cancellation of:

   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).

   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

* *If item 4 applies, some or all of these sheets may be marked "superseded."*

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/EP2005/050739

Box No. IV Lack of unity of invention

1. ☒ In response to the invitation to restrict or pay additional fees, the applicant has, within the applicable time limit:

   ☐ restricted the claims.

   ☒ paid additional fees.

   ☐ paid additional fees under protest and, where applicable, the protest fee.

   ☐ paid additional fees under protest but the applicable protest fee was not paid.

   ☐ neither restricted the claims nor paid additional fees.

2. ☐ This Authority found that the requirement of unity of invention is not complied with and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is:

   ☐ complied with.

   ☒ not complied with for the following reasons:

   see separate sheet

4. Consequently, this report has been established in respect of the following parts of the international application:

   ☒ all parts.

   ☐ the parts relating to claims Nos. .

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-32
	No:	Claims	
Inventive step (IS)	Yes:	Claims	1-32
	No:	Claims	
Industrial applicability (IA)	Yes:	Claims	1-32
	No:	Claims	

2. Citations and explanations (Rule 70.7):

   see separate sheet

Supplemental Box relating to Sequence Listing

Continuation of Box I, item 2:

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, this report was established on the basis of:

   a. type of material:

   ☒ a sequence listing

   ☐ table(s) related to the sequence listing

   b. format of material:

   ☒ on paper

   ☒ in electronic form

   c. time of filing/furnishing:

   ☒ contained in the international application as filed

   ☒ filed together with the international application in electronic form

   ☐ furnished subsequently to this Authority for the purposes of search and/or examination

   ☐ received by this Authority as an amendment* on

2. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

* *If item 4 in Box No. I applies, the listing and/or table(s) related thereto, which form part of the basis of the report, may be marked "superseded."*

Citations

The documents mentioned in this International Preliminary Report on Patentability (IPRP) are numbered as in the International Search Report (ISR) dated27.10.05, i.e. **D1** and **D7** correspond to the first and the last document of the search report, respectively. The ISR has been established by this authority.

Re ITEM IV (Unity of invention)

1 In response to an invitation, the Applicant paid two additional search and two additional examination fees. Consequently, international search and examination have been carried out for the subject-matter of claims 1-32 (inventions 1-3). The present application lacks unity as required by Art. 3(4)(iii) and Rule 13 PCT because it contains 3 seperate inventions:

1.1 Invention 1: claims 1-18
A method for the purification of a virus comprising adding a nuclease to host cells that are infected with a virus before lysing or before 95% of the host cells have been lysed by a virus capable of lysing host cells, respectively.

1.2 Invention 2: claims 19-29
A method for the production of a virus comprising a nucleic acid sequence coding for a nucloeprotein of a heamorrhagic fever virus, comprising culturing host cells that have been infected with said virus, lysis of the host cells and subjecting the virus to anion exchange chromatography.

1.3 Invention 3: claims 30-32
A method for removing free adenovirus proteins from a recombinant adenovirus preparation, comprising the step of subjecting a recombinant adenovirus preparation comprising free adenovirus proteins to a charged filter that contains anion exchange groups.

2 According to Art. 3(4)(iii) and Rule 13 PCT an application shall relate to one invention only or to a group of inventions so linked as to form a single general inventive

concept. Where a group of inventions is claimed, the requirement of unity of invention referred to in Rule 13.1 PCT shall be fulfilled only when there is a technical relationship among those inventions involving one or more of the same or corresponding special technical features.

3. The special technical feature of invention 1 is the addition of nuclease to a culture of host cells that are infected with a virus before lysing said host cells or before complete lysis of the host cells by a virus capable of lysing host cells, respectively.

4 In the methods of inventions 2 and 3 no nuclease at all is required. Invention 3 is not concerned with viruses comprising a nucleic acid sequence coding for a nucleoprotein of a heamorrhagic fever virus.

Since none of inventions 2 and 3 share the special technical feature of invention 1 and since no other technical feature can be distinguished which might link any of inventions 1-3, each of the above mentioned groups of claims represents an independent invention.

5 In view of the above the only "single general concept" (Rule 13.1 PCT) linking the above mentioned inventions can be formulated as methods for the purification of a virus or purified virus, respectively. This concept is, however, not novel with regard to the prior art:

D3 (WO03097797), for instance, discloses methods of adenovirus purification wherein contaminating host cell DNA levels are reduced to less than 5 pg/10^{11} vp.

6 Because said single general concept is evidently not novel it cannot be inventive as required by Rule 13.1 PCT.

N.B.: The use of the term "invention" here in no way implies recognition of an inventive step for the subject-matter of any group of claims.

Re ITEM V (Novelty, inventive step, industrial applicability)

1 Novelty (Art. 33(2) PCT)

invention 1:

1.1 The subject-matter of claims 1-18 has not been made available to the public by any of the available prior art documents and can therefore be regarded as novel.

invention 2:

1.2 The subject-matter of claim 19-29 has not been made available to the public by any of the available prior art documents and can therefore be regarded as novel.

invention 3:

1.3 The subject-matter of claim 30-32 has not been made available to the public by any of the available prior art documents and can therefore be regarded as novel.

2 Inventive step (Art. 33(3) PCT)

invention 1:

2.1 The subject-matter of claim 1-18 cannot be derived from the available prior art in an obvious manner and therefore complies with the requirements of Art. 33(3) PCT.

2.2 **D1** (Drittanti et al.), **D2** (WO9822588) and **D3** disclose a method comprising the steps a, b and c (claim 1) in the order a, c, b. Thus, in the prior art methods of purifying viruses nuclease is added **after** complete lysis of the host cells. Adding nuclease before lysis or before lysis has completed, respectively, is not suggested or layed near in the available prior art.

invention 2:

2.3 The subject-matter of claim 19-29 cannot be derived from the available prior art in an obvious manner and therefore complies with the requirements of Art. 33(3) PCT.

2.4 The prior art discloses adonviruses comprising a nucleic acid sequence coding for an Ebolavirus nucleoprotein (NP) (see e.g. **D7** (Sullivan et al., abstract and Methods)). Methods for the production of viruses comprising a nucleic acid sequence coding for

a nucleic acid binding protein are also known from the prior art (see e.g. **D4** (US20020182723), **D5** (US6261823) or **D6** (Green et al.)).

2.5 However, a method for the production of a virus comprising a nucleic acid sequence coding for a nucleoprotein of a heamorrhagic fever virus is not obvious in view of the available prior art.

invention 3:

2.6 The subject-matter of claim 30-32 cannot be derived from the available prior art in an obvious manner and therefore complies with the requirements of Art. 33(3) PCT.

2.7 According to p. 27, last line-p. 28, l. 9 of present application "...certain adenovirus proteins that were not incorporated into adenovirus particles are separated form the AV particles by use of an anion exchange filter, not by an anion exchange column. Such free AV proteins were not previously found in preparations of recombinant AV particles and would normally go undetected, but now can be removed using the step of subjecting a recombinant AV preparation comprising free AV proteins to a charged filter that contains anion exchange groups". **D3** discloses methods for the purification of adenoviral (AV) preparations. **D3** mentions the use of anion exchange membrane chromatography (p. 24, l. 24-26). However, **D3** does not mention or suggest the purpose of such use as defined in present independent claim 30, namely the "removal of free AV proteins". Thus, a method for removing free AV proteins according to claim 30 cannot be regarded as obvious.

3 Industrial application (Art. 33(4) PCT)

Claims 1-32 meet the criteria as set forth by Art. 33(4) PCT.